96-WELL HOLOLID™

PRODUCT DESCRIPTION AND INSTRUCTIONS

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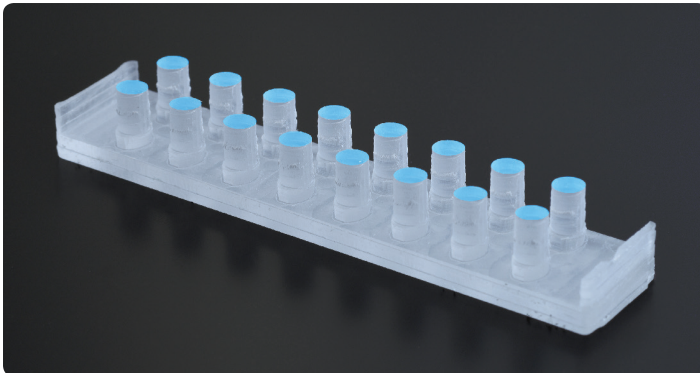
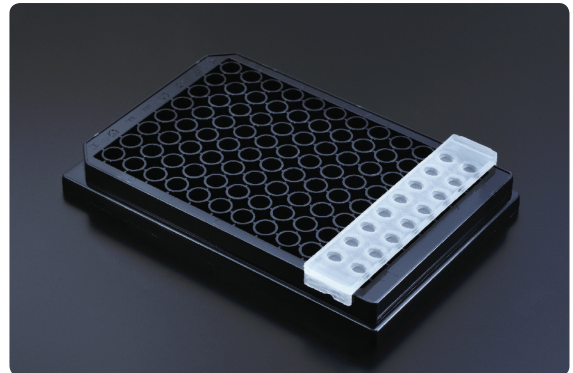
Phase Holographic Imaging

The PHI 8060 96-well HoloLid has been especially designed for the HoloMonitor® time-lapse cytometers to eliminate image disturbances caused by surface vibrations and condensation inside the cell culture vessel. The lid is designed to fit Sarstedt 96-well, lumox® base plates with an ultra-thin gas permeable membrane which allows for a good gas exchange (Sarstedt Cat. # 94.6000.024). As no further ventilation is necessary the size of the imaging area has been maximized.

The lid needs to be sterilized before use and can be reused at least 10 times. However, after extensive use the repeated sterilization will noticeably degrade the optical quality of the lid.





96-well HoloLid, bottom facing up. The blue areas are the surfaces that are immersed into the cell media (left). 96-well HoloLid placed in a Sarstedt 96-well plate (right).

FORMAT

85.6 × 17.6 × 11.9 mm (exterior) and ø4.2 mm (observation window). Each lid covers two columns (16 wells). To cover an entire plate 6 lids are needed.

MATERIAL

Poly methyl methacrylate (PMMA, Plexiglas), a non-toxic material often used in medical surgery implants, dentures etc. It does not contain Bisphenol-A, a cell disturbing agent commonly present in plastics. The lid is shipped with a plastic cover that must be peeled off before the first usage. The lid is reusable and has to be sterilized before use.

STERILIZING

1. Place the PHI 96-well HoloLid into a cleansing bath with warm water and detergent for at least 10 minutes.
2. Rinse in multiple steps with tap water first and ultra-pure water last.
3. Place the lid into a bath with 70 % non-denatured ethanol inside the sterile bench for 15 minutes. It is very important to keep the ethanol bath as short as possible as ethanol affects the optical quality of the plastic. Handle the lid with sterile tweezers and store in a sterile fashion until used, a square Petri dish of 100 × 100 mm is recommended.

USAGE

All steps below are to be handled with standard sterile procedures.

1. Seed the cells. A working volume of 170 µl for each well is recommended (adjusted to reach a surface level that allows the observational window to be immersed). Put on the normal lid.
2. Let the cells adhere in the incubator for 1-5 hours, depending on the required adherence time for the specific cells used. This step is performed to avoid uneven distribution of cells. If a reagent is to be added one day after seeding, it is recommended to change lids after the addition.
3. Replace the normal multi-well plate lid with the 96-well HoloLid. Make sure there are no air bubbles in the cell media before changing the lids. If there is an air bubble it can easily be removed by blowing a little puff of clean air onto the bubble which will burst. Clean air can be created e.g. by using an ethanol dispensing bottle with a little ethanol inside and the inner tube removed. Press carefully the bottle while targeting the bubble with its tip.
4. The sample is ready to be used.

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